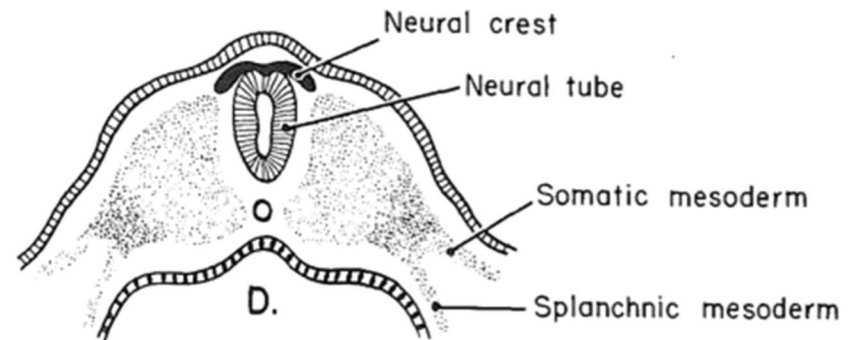
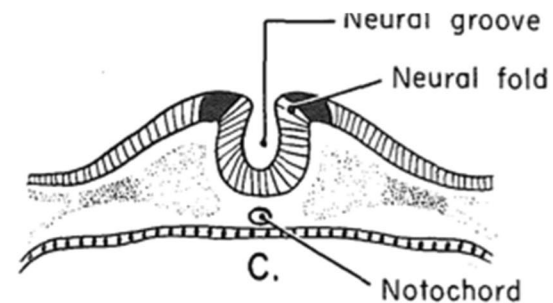
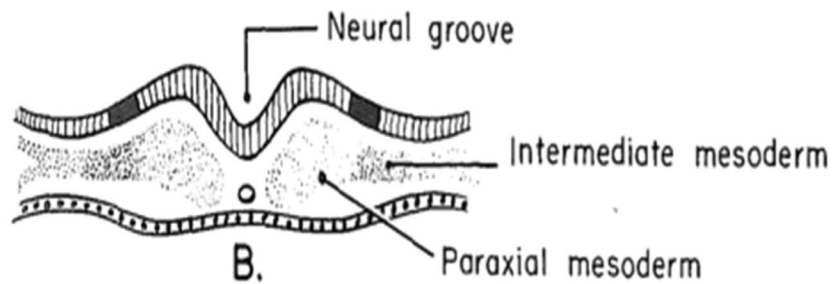
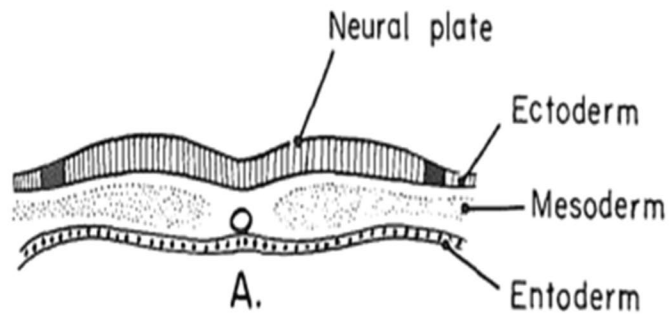


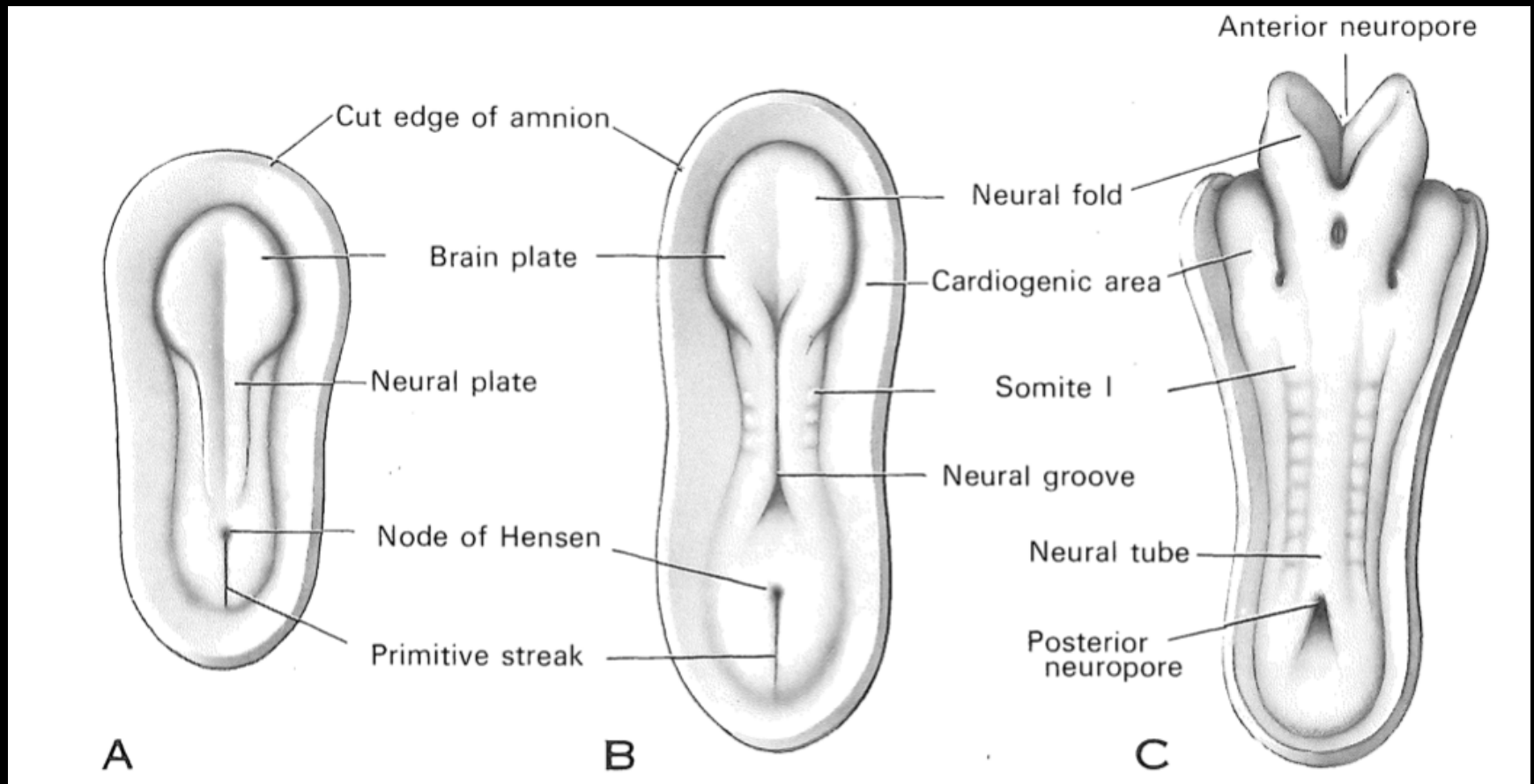
# Dr. Hevner's Lab

Lauren Honican and Olachi Anamelechi  
UW Neurological Surgery Summer  
Student Program

# Histogenesis of the Nervous System



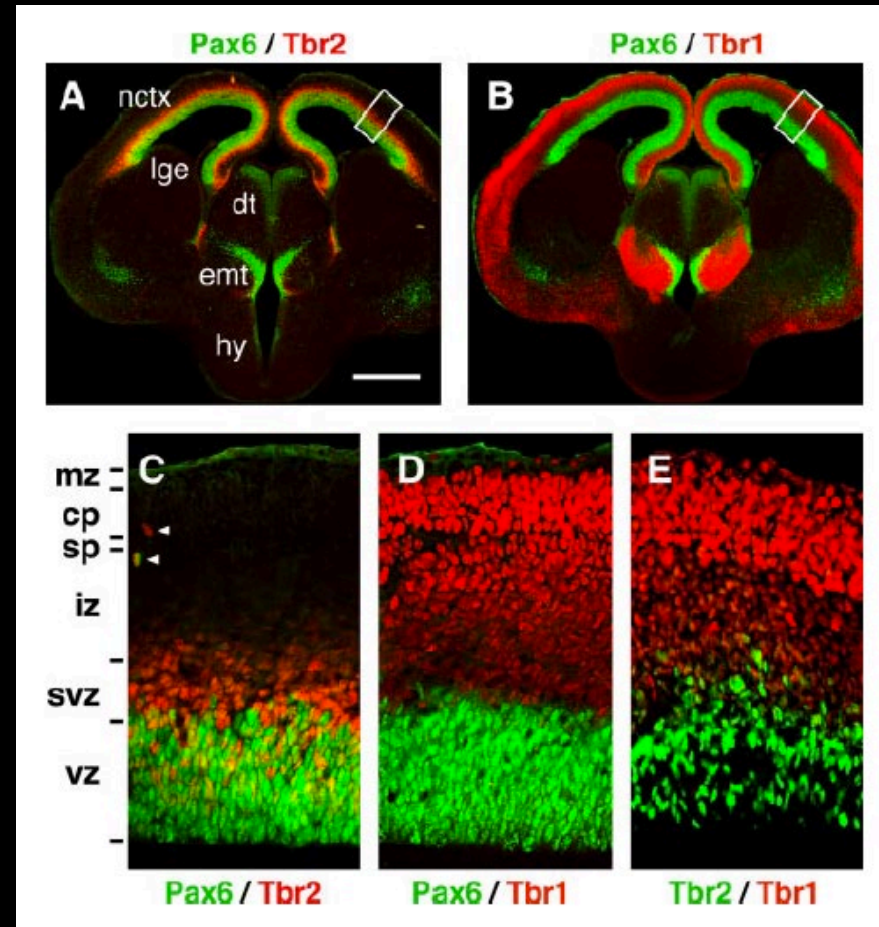
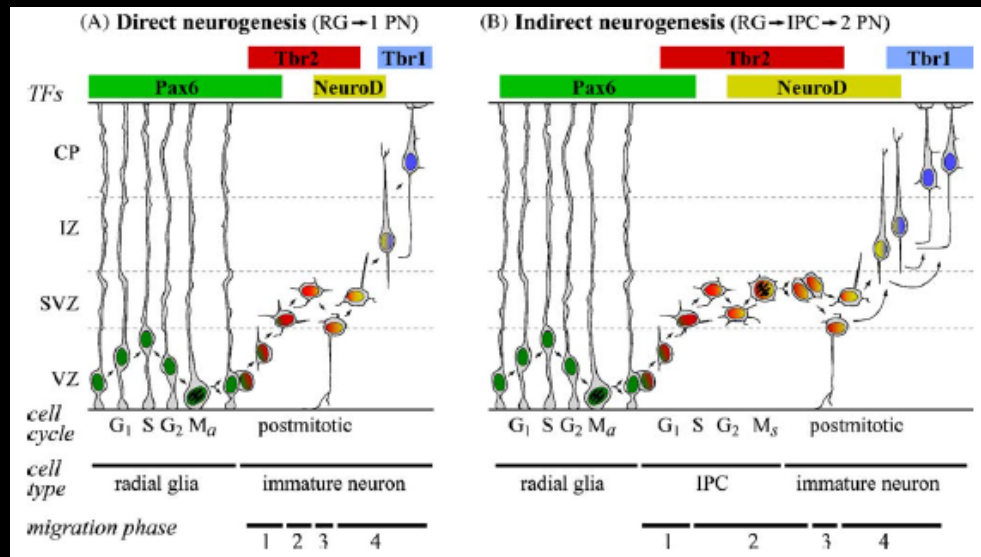
# Formation of the Nervous System






# Neurodevelopment of embryonic mouse cortex:

## Transcription factor (TF) cascades

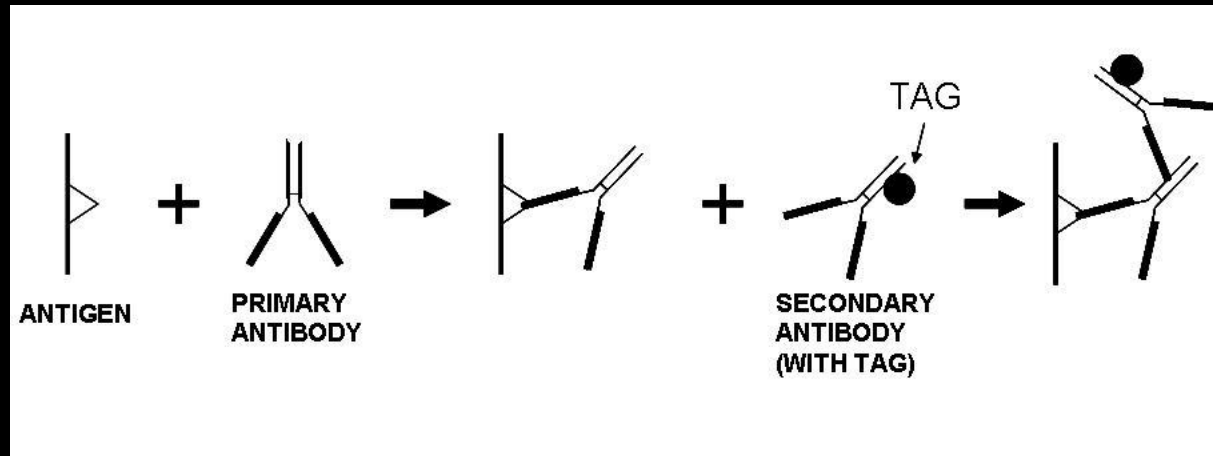




# Pituitary Adenylate Cyclase-Activating Polypeptide (PACAP)

- Neurotransmitter that acts primarily on PAC1 receptors
- Vital to pro and anti-mitogenesis, and postnatal survival.
- Serves as a precursor to proliferation, differentiation, and final cell population size.
- Evidence Links PAC 1 short isoforms to anti-mitogenesis in E13.5 and later, while PACAP hop isoforms serve to promote proliferation in early corticogenesis.

# Immunohistochemistry



- Antigen (antibody generator)
  - Promotes an immune response (the secretion of antibodies) within the body.
  - Antibodies bind to epitopes on the surface of antigens.
- Primary antibody
  - Antigen-specific.
  - Produced by B-cells (lymphocytes).
- Fluorescent secondary antibody
- DAPI counterstain

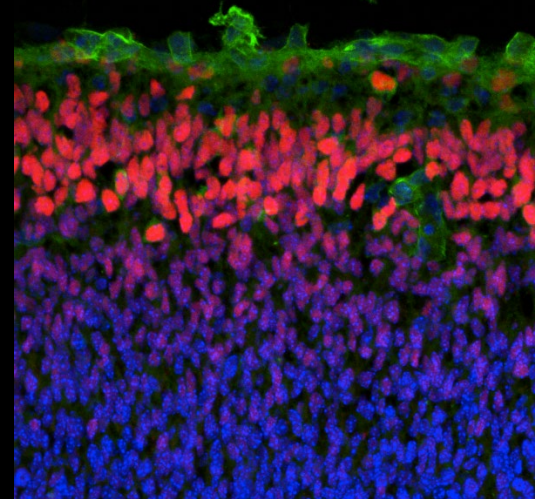


# Methods

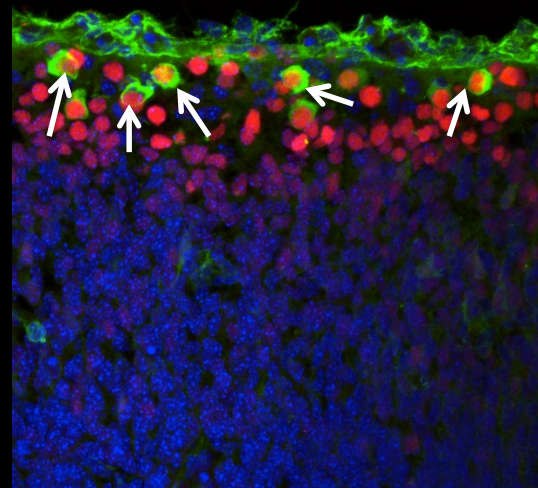
- Experiment 1:
  - Stained E14.5 sagittal sections of WT and Tbr2cko mice with PACAP
  - Examined under fluorescence microscope.
- Experiment 2:
  - Double stained E14.5 sagittal sections of WT and Tbr2cko mice with PACAP and Tbr1
  - Examined using fluorescence microscope and confocal.

# Immunofluorescent imaging results

- Experiment 1: E14.5
  - WT: Low PACAP expression
  - Tbr2cko: PACAP highly expressed in CP
- Experiment 2: E14.5
  - WT: Tbr1 expressed in deep cerebellar nuclei, CP, DG and OB. Low PACAP expression.
  - Tbr2cko: PACAP expressed in developing hippocampus in DG and CP. Co-localization of Tbr1 and PACAP.



Control



Tbr2cko

Color key: DAPI, Tbr1, PACAP



# Conclusions

- Experiment 1:
  - PACAP is upregulated in Tbr2cko
  - Tbr2, TF in intermediate zone (IZ), may act indirectly to inhibit PACAP expression
- Experiment 2:
  - Tbr1, TF in cortical plate (CP), is a marker of post-mitotic neurons.
  - Co-localization demonstrates that PACAP cells are post-mitotic neurons.

# Further research

- What specific type of neurons are PACAP cells?
- How does PACAP influence regulation of neural development?
- What role does PACAP play in development of the hippocampus?



# Many thanks

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- Ray Daza
- Kevyn Ramos
- Christina Buckman
- Jim Pridgeon

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